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ORIGINAL ARTICLE

Efficacy of THN102 (a combination of modafinil and flecainide) on vigilance and cognition during 40-hour total sleep deprivation in healthy subjects: Glial connexins as a therapeutic target

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Aims: THN102 is a novel combination of modafinil and low-dose flecainide, targeting glial connexin activity to modulate modafinil effects. We investigated THN102 efficacy compared to modafinil and to placebo on vigilance and cognitive function during 40-hour total sleep deprivation (TSD).

Methods: Twenty healthy men participated in a double-blind, randomized, incomplete-block 3-period cross-over trial with 5 treatments (n = 12 per group): placebo (PBO), modafinil 100 mg (MOD100), THN102 100/1, 100/3, 100/9 (modafinil 100 mg and flecainide 1, 3 or 9 mg). Each period included a baseline day and a TSD day with treatments administered 3 times (01:00, 09:00 and 19:00). Reaction time in psychomotor vigilance test, subjective somnolence and vital signs were assessed before and during treatment. Working memory (2-Back) and executive processes (Go/noGo for vigilance and inhibition, Wisconsin card sorting task for mental flexibility, and Tower of London test for planning) were evaluated at 16:30.

Results: At 5 hours postdose−1 (after 23 hours TSD, primary endpoint), THN102 100/1 resulted in statistically higher psychomotor vigilance test speed *vs* MOD100 (3.97 ± 0.09 *vs* 3.74 ± 0.14, $P < .05$). No increase in effect was observed with higher flecainide doses in combinations. Most THN102 doses *vs* MOD100 also improved the number of correct responses in 2-Back and Go errors in Go/noGo ($P < .05$ for all doses), and perseverative responses in Wisconsin card sorting task (for 100/1 and 100/9). No impact on cardiac conduction was noted with THN102, and safety was similar to MOD100.

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The authors confirm that the Principal Investigator for this paper is Dr Fabien SAUVET and he had direct clinical responsibility for patients.
Clinical trials: name, URL, and registration: NCT 03182413, Impact of THN102 on Attention, Wakefulness and Cognitive Performance During Total Sleep Deprivation.

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Conclusions: THN102 seems more efficient than modafinil on vigilance, working memory and executive functions, opening new perspectives in management of hypersomnolence disorders.

KEYWORDS

executive function, flecainide, modafinil, psychomotor vigilance task, safety, sleep deprivation, sustained attention, working memory

1 | INTRODUCTION

Modafinil is a nonamphetaminic wake-promoting compound indicated in the USA for the treatment of excessive daytime sleepiness (EDS) in narcolepsy,[1,2] obstructive sleep apnea[3] and shift work disorder.[4] Additionally, modafinil has been recommended to alleviate EDS in Parkinson's disease[5] and during prolonged military operations for vigilance and cognitive performance.[6-10] Modafinil has demonstrated efficacy on vigilance, attention, executive processes and memory during experimental protocols of sleep deprivation.[11-13] However, it is estimated that 50–70% of the narcoleptic patients still experience EDS despite psychostimulants and notably modafinil use.[14,15] Meanwhile, narcoleptic patients complain of attention deficit, with altered executive processes and memory, despite treatments.[6,16]

The wake-promoting mechanism of action of modafinil remains unclear, as it has been proposed to act on several neuronal circuits, including through increases in dopamine levels (inhibition of dopamine transporter).[17] Moreover, modafinil has been recently demonstrated to act as a cellular-coupling enhancer in astrocytes, through the modulation of gap junctions constituted by connexins.[18] In particular, astroglial connexins are involved in sleep and homeostasis regulation,[18,19] as sleep deprivation and subsequent sleep rebound modify their expression and their inhibition causes sleep loss,[20,21] and are more largely involved in neuronal modulation.[22] In addition, an astroglial connexin modulator, flecainide,[23] has been shown to significantly enhance the wake-promoting and procognitive effects of modafinil in nonclinical models.[24] Modafinil effects on astroglial cell coupling mediated by connexin 30 were reversed by flecainide.[24] Altogether, these results indicate that low-dose flecainide improved the wake-promoting and procognitive functions of modafinil, probably through the changes of connexin-dependent gap junctional coupling in astroglial networks. Non-clinical studies in rat indicated that THN102 significantly increased regional brain glucose metabolism in the cortex, striatum and amygdala compared to control or drugs administered alone.[25]

The present study aims to evaluate the effect of the combination of modafinil and flecainide low dose, namely THN102, as a clinical proof of concept of its efficacy on vigilance and cognition. The primary objective was to investigate the efficacy of THN102 (modafinil 100 mg + flecainide low dose 1, 3 or 9 mg) compared to modafinil alone for improving sustained attention during a 40-hour total sleep deprivation (TSD) protocol in healthy subjects, using the psychomotor vigilance test (PVT). Secondary objectives were to assess safety and impact of treatment with THN102 on working memory and executive processes compared to placebo and modafinil alone.

2 | METHODS

2.1 | Subjects

Twenty healthy male subjects, age range 20–40 years, with body mass index <30 kg/m^2, were included in the study. Subjects were screened with their medical and psychiatric history, they underwent a physical examination, electrocardiography evaluations and laboratory tests (haematology, clinical chemistry, urinalysis). Subjects were not shift workers and had not travelled between time zones within 14 days prior to the study.

Exclusion criteria included physical or mental health troubles based on: (i) Hospital Anxiety and Depression scale ≥16[26]; (ii) significant medical history; (iii) Epworth Sleepiness Scale, >11[27]; (iv) Pittsburgh sleep quality index >8; (v) Horne & Östberg morningness–eveningness questionnaire <31 or > 69[28]; (vi) habitual time in bed per night <6 hours.

The study received the agreement of the Cochin—CPP Ile de France 1 (Paris) Ethics Committee and of the French National Agency for Medicines and Health Products Safety (EudraCT 2015–001927-21, NCT03182413). It was conducted according to the principles expressed in the Declaration of Helsinki of 1975, as revised in 2001 after obtaining written informed consent from all the subjects.

2.1.1 | Study design

This was a single centre, double-blind, randomized, incomplete-block cross-over trial, placebo and modafinil-controlled study, with 5 treatments given over 3 periods in the sleep laboratory in Hospital Percy (Clamart, France), with a 4-week washout between periods (see Figure 1). Hence, every participant only takes part in 3 of 5 possible conditions, during Periods I, II and III. The cross-over design selected for this study was design No. 8,[29] which allows balancing the sequence between the subjects (1 sequence per subject). This is a variance-balanced design, meaning that all pairwise treatment comparisons will be performed with the same precision. Each treatment is given to 12 subjects. Compared to the gold standard consisting of a full 5-treatment–5-period cross-over design involving 2 orthogonal Latin squares, the efficiency of the design for the direct treatment

comparison remains strong (83%). It is also suitable for detecting treatment effects in the presence of a carry-over (68%).[30] The 5 conditions of treatments were: PBO (placebo_modafinil and placebo_flecainide), MOD100 (modafinil 100 mg with placebo_flecainide), THN102 100/1 (modafinil 100 mg + 1 mg flecainide), THN102 100/3 (modafinil 100 mg + 3 mg flecainide), THN102 100/9 (modafinil 100 mg + 9 mg flecainide). Each subject received 3 of the 5 possible treatments according to a unique sequence (one sequence per subject; Table 1). Treatments were administered 3 times during each period; allocation was balanced. Key protein targets and ligants in the article are hyperlinked to corresponding entries in the http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS guide to pharmacology.

All substances were administered double-blind so that neither the volunteers nor the investigators nor sponsor were aware of the conditions until database lock after the completion of the final run of the study and final data review. An emergency procedure of unblinding had been created for a potential major undesirable effect.

Each study period included a 4-day hospitalization (see Table 2): (i) habituation/training day (D −1); (ii) baseline day (D0); (iii) TSD for 40 hours beginning on D0 at 7:00 until D1 at 23:00 and treatment day with 3 oral doses at 01:00, 09:00 and 19:00 (D1); (iv) recovery day starting after a night of sleep (D2).

An end-of-study visit was planned 8 days after the end of the last study period (Period III) with complete physical examination and laboratory safety.

2.1.2 | Procedures

The PVT (speed), vital signs (sitting blood pressure, heart rate), visual analogue scale (VAS) for somnolence and intensity of key gastrointestinal and central nervous system symptoms, blood pressure and electrocardiography (ECG) were assessed at baseline (D0), before first dose, and 9 times after first dose. Executive function parameters were compared at similar timing (i.e. at 16:30 during D1 [15.5 hours after the beginning of treatment] *vs* D0). Executive function tests included working memory (2-back), planning (Tower of London, ToL), mental flexibility (Wisconsin card sorting test, WCST) and inhibition (Go/noGo).

During all the periods in the sleep laboratory, including the tests, electroencephalogram, electrooculography, electromyography and ECG were continuously recorded (Actiwave, CamNtech Ltd England). Polysomnography data were scored by 2 trained technicians in accordance with AASM Manual for the Scoring of Sleep and Associated Events (2007) criteria using Somnologica software (Medcare, Reykjavik, Iceland) to confirm that all subject stayed awake during the 40-hours continuous wakefulness period (see Table 2).

2.1.3 | Measurements

PVT

We utilized a computer-based version of the 10-minute PVT.[31] Subjects were instructed to monitor a red rectangular box on the computer screen and press a response button as soon as a yellow stimulus counter appeared on the screen, which displayed the reaction time (RT) in milliseconds for a 1-second period. The interstimulus interval, defined as the period between the last response and the appearance of the next stimulus, varied randomly from 2 to 10 seconds. PVT response was regarded valid if RT was ≥100 ms. The PVT speed was expressed as (1/reaction time × 1000). The ratios of speeds (each timepoint over timepoint 0; Baseline) for THN *vs* MOD and *vs* PBO treatments were statistically analysed. Comparison between mean reaction speeds (mean of 1/RTs) was considered as primary endpoint of this study at 5 hours postdose 1; mean speed was also assessed at 11 other timepoints.[32]

What is already known about this subject

- THN102 is a combination of modafinil and low-dose flecainide targeting neuroglial interaction.
- It improved wakefulness and cognition compared to modafinil alone in animal models.

What this study adds

- THN102 is superior to modafinil alone and placebo in vigilance performance in sleep-deprived healthy men.
- THN102 improves working memory and executive functions in sleep-deprived healthy men.
- The enhancement of alertness and cognitive outcomes with THN102 opens new clinical perspectives.
- THN102 may have clinical interest in improving attention and cognitive deficits associated with hypersomnolence disorders.

Executive function tests

Tests were computed on PEBL test Battery.[33]

Working memory was assessed using a 10-minute visual 2-back task.[34] In this task, the subject has to constantly keep in memory the last 2 letters of a random sequence. One new random *n* letter of the sequence was presented to the subject every 4 seconds and the subject has to respond should this letter correspond to the *n*−2 letter, with a requirement for rapid responses while maintaining accuracy. Percentage of correct responses and mean response times were computed.

In the inattention and impulsivity Go/noGo task[35] subjects were required to watch a sequential presentation of letters and respond to a target letter by pressing a button. A single letter (P or R) was presented for a duration of 500 ms with an interstimulus interval of 1500 ms. In the first condition (P-Go), participants had to press a button in response to the target letter P and withhold their response to the nontarget letter R. The first condition consisted of 160 trials. A second, reversal condition (R-Go), was then administered, and participants were required to make a response to the target letter R and

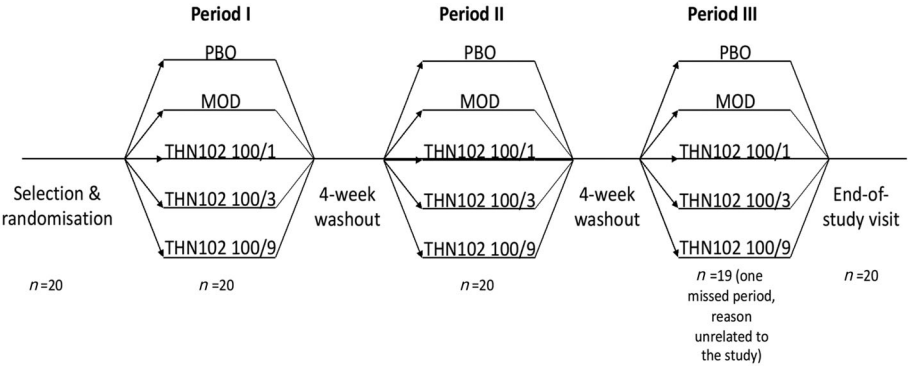

FIGURE 1 Trial design. Treatments were composed of placebo (PBO, placebo_modafinil and placebo_flecainide), modafinil 100 mg with placebo_flecainide (MOD100) or with flecainide 1, 3 or 9 mg (THN102 100/1, THN102 100/3, THN102 100/9)

withhold their response to the nontarget letter P. Ratios of targets to nontargets were 80:20 for both sequences. Performance of the task was assessed by calculating errors of omission the Go letter and errors of commission in noGo sequence.[35]

The ToL test allows to assess planning aspects of executive function.[36] Participants completed a 10-trial computerized version. Each trial presented volunteers with a series of 3 *pegs* of differing lengths, each with 3 different coloured *beads* arranged in randomly appearing configurations upon the pegs. The examinee was required to rearrange the beads so that the final configuration matched a prespecified goal pattern. The goal was to complete the task in as few moves and as quickly as possible. Dependent variables from this task included the number of moves required to match the goal arrangement, and the mean pickup time (ms; time to solve the problem).

Subjects also performed the WCST to assess flexibility in executive function.[37] The computer screen presented a deck of cards and 4 key cards. The key cards each displayed a different design (4 cards with 1–4 shapes in 4 different colours). The card deck contained 64 cards of varying shapes, colours and numbers of shapes. Participants were told to match each card from the deck to 1 of the 4 key cards. Once an option was selected, the computer moved the card to a position just below the associated key card. Without warning, the sorting principle changed after the examinee had completed 10 consecutive correct matches. The total number of errors, perseverative responses (responses that follow a previously reinforced principle that is no longer correct) and perseverative errors (i.e. errors that follow a previously reinforced principle that is no longer correct), were calculated.

TABLE 1 Treatment sequences in the 3-period trial

Subject	P1	P2	P3
S111	MOD100	PBO	THN 100/1
S115	MOD100	THN 100/1	PBO
S116	MOD100	THN 100/3	THN 100/9
S105	MOD100	THN 100/9	THN 100/3
S103	PBO	MOD100	THN 100/9
S110	PBO	THN 100/1	THN 100/3
S101	PBO	THN 100/3	THN 100/1
S107	PBO	THN 100/9	MOD100
S113	THN 100/1	MOD100	THN 100/3
S102	THN 100/1	PBO	THN 100/9
S106	THN 100/1	THN 100/3	MOD100
S108	THN 100/1	THN 100/9	PBO
S118	THN 100/3	MOD100	PBO
S104	THN 100/3	PBO	MOD100
S119	THN 100/3	THN 100/1	*THN 100/9**
S112	THN 100/3	THN 100/9	THN 100/1
S109	THN 100/9	MOD100	THN 100/1
S117	THN 100/9	PBO	THN 100/3
S114	THN 100/9	THN1	MOD100
S120	THN 100/9	THN3	PBO

*missing period.

2.1.4 | VAS

In addition to spontaneously reported adverse events (AEs) the intensity of 6 symptoms frequently associated with the use of modafinil[12,38]—headache, nausea, abdominal pain, dry mouth, somnolence and fatigue—were evaluated using a 100 mm VAS. Intensity above 20 mm was considered as AE pattern. If observed after the treatment administration, AE was considered as related to treatment.

2.2 | Statistical analysis

Statistical analyses were computed using SAS for the primary endpoint and adverse events, and using R (version 3.3.1; 2016-06-21) for other parameters. Values were expressed as mean ± standard error.

The primary objective was to demonstrate the superiority of THN102 *vs* MOD100 at the nadir time point (at 06:00 on Day 1, 5 hours after dose 1). The 06:00 hour corresponds to the highest decrease of attentional performance, observed after 23 hours of sleep deprivation, due to continuous awakening (sleep pressure) and the nadir of circadian rhythm.[11,12,39,40] With 20 evaluable subjects and at least 57 evaluable periods (i.e. a maximum of 3 missed periods), the study has at least 80% power to detect a significant difference in the primary endpoint (mean difference in PVT speed at nadir) when

TABLE 2 Time frame for each period

Day	Time of day	Cumulative sleep deprivation (h)	Relatives hours before and after dose 1 (h)	Events
-1	13:00	-	-	Arrival in the laboratory, medical examination
-1	23:00	-	-	Lights off—Baseline sleep (8 h in bed)
0	07:00	0	−18	Awake
0	10:00	3	−15	PVT, VAS, BP, ECG
0	16:30	9.5	−8.5	2-Back, ToL, WCST, go/noGo **(baseline)**
0	23:30	16.5	−1	PVT, VAS, BP, ECG **(baseline)**
1	01:00	18	0	**Drug or placebo (dose 1)**
1	03:30	20.5	2.5	PVT, VAS, BP, ECG
1	06:00	23	5	PVT **(primary endpoint),** VAS, BP, ECG
1	08:00	25	7	PVT, VAS, BP, ECG
1	09:00	26	8	**Drug or placebo (dose 2)**
1	10:00	27	9	PVT, VAS, BP, ECG
1	12:30	29.5	11.5	PVT, VAS, BP, ECG
1	15:30	32.5	14.5	PVT, VAS, BP, ECG
1	16:30	33.5	15.5	2-Back, ToL, WCST, go/noGo
1	18:00	35	17	PVT, VAS, BP, ECG
1	19:00	36	18	**Drug or placebo (dose 3)**
1	21:00	38	20	PVT, VAS, BP, ECG
1	23:00	40	22	Lights off—Recovery sleep (8 h in bed)
2	07:00	-	30	Awake
2	10:00	-	33	PVT, VAS, BP, ECG
2	11:30	-	34.5	Discharge

PVT, psychomotor vigilance test; VAS, visual analogy scale (tolerance); BP, (blood pressure); ECG, electrocardiogram. Executive function tasks are: ToL, Tower of London test (executive function); WCST, Wisconsin card sorting task (mental flexibility); Go/noGo, Go/noGo task (mental inhibition); 2-Back, 2-Back memory task.

comparisons are made at the unadjusted 2-sided 5% level. Powering was made assuming an effect size equal to 1.3 and a true within-subject standard error of 37.9 ms in reaction time in the PVT, as reported for placebo treatment in our laboratory.[39,40] PVT mean speed was analysed at each time point separately using a mixed-effect analysis of variance (ANOVA) model including fixed effects for treatment and period, a random effect for subjects, and baseline value (i.e. value observed at 23:30, D0) as a covariate (posthoc analysis).[41-43] As additional analysis, area under the curve (AUC) for PVT speed was calculated using the classical linear trapezoidal approach, for the first 9 and 20 hours of TSD (AUC_{0-9}, AUC_{0-20}). The same statistical model as described above was used to analyse secondary endpoints (executive function tests, safety). Descriptive statistics were calculated for line analogue rating scales and for each treatment and time point. Data are available on request from the corresponding author.

3 | RESULTS

Subjects

Twenty-three subjects were screened and 20 were included and randomized, with a mean age of 28.9 ± 5.6 years (range: 21–39 years), a mean body weight of 72.3 ± 8.5 kg (range: 55–92 kg) and body mass index of 23.1 ± 1.85 kg/m^2 (range: 19.5–26.2 kg/m^2). One subject (subject 119) participated in only 2 periods (reason unrelated to the study) but was not considered as a drop out as he completed his end-of-study visit. Overall, no drop out was observed. The emergency unblinding procedure has never been used.

PVT speed, primary endpoint

No differences between treatments were observed at the baseline point (23:30 in D0) and no differences between baselines of the 3 periods (absence of carryover). In the PBO group, PVT speed rapidly decreased to early morning nadir observed at 06:00. During sleep deprivation, we observed a higher PVT speed for THN102 100/1 *vs* MOD100 at 5 hours postdose 1 (i.e. at 06:00, $P < .05$; Table 3; Figure 2A).

Moreover, after THN102 100/1 treatment, PVT speed was higher compared to PBO in 6 time-points (2.5, 5, 7, 11.5, 14.5 and 20 hours postdose 1), while MOD100 was different from PBO at only 3 time-points (5, 11.5 and 20 hours; Table 3).

Differences between THN102 doses (1, 3 or 9 mg of flecainide) were not observed, at any time point, as well as no ordinal trend effect. Overall, the results of the THN102 doses show a consistent pattern above the performance with modafinil alone (Table 3).

Globally, THN102 100/1 was more efficient on PVT speed compared to MOD100 during the first 9 hours of treatment (AUC_{0-9}, $P < .05$; Figure 2B).

Executive functions

MOD100 during sleep deprivation (D1) failed to restore basal level (D0) in working memory (2-Back, Figure 3), mental flexibility (WCST, Figure 4) and mental inhibition (Go/NoGo, Figure 4) *vs* PBO. Interestingly, THN102 demonstrated significant improvements *vs* MOD100 and *vs* PBO in those processes (Figures 3 and 4). In particular, after all THN102 doses we observed fewer GO errors and higher correct responses in the 2-Back test ($P < .05$ for all). Perseverative errors in the WCST were lower *vs* MOD100 and *vs* PBO for THN102 100/1

TABLE 3 Comparisons between treatments for PVT mean speed (1/reaction time)— pharmacodynamic (PD) analysis set

Relative hours before and after dose 1 (h)	Mean contrast with placebo (95%CI)/*P*-value				Mean contrasts with modafinil (95% CI)/*P*-value		
	THN102 100/9 - PBO	THN102 100/3 - PBO	THN102 100/1 - PBO	MOD100 - PBO	THN102 100/9 - MOD100	THN102 100/3 - MOD100	THN102 100/1 - MOD100
−15 h	1.01 (0.97–1.06)	0.99 (0.94–1.03)	0.98 (0.94–1.03)	0.98 (0.94–1.03)	1.03 (0.98–1.08)	1.00 (0.96–1.05)	1.00 (0.96–1.05)
0 (baseline)	1.03 (0.98–1.08)	1.01 (0.96–1.06)	1.00 (0.96–1.05)	1.00 (0.96–1.05)	1.03 (0.98–1.07)	1.00 (0.96–1.05)	1.00 (0.95–1.04)
2.5 h	1.05 (0.99–1.12)	1.04 (0.98–1.11)	**1.08 (1.02–1.15)***	1.03 (0.97–1.10)	1.02 (0.96–1.09)	1.01 (0.95–1.07)	1.05 (0.90–1.11)
5 h (primary endpoint)	**1.06 (1.01–1.11)***	**1.05 (1.01–1.10)***	**1.11 (1.06–1.16)***	**1.06 (1.02–1.11)***	1.00 (0.96–1.04)	0.99 (0.95–1.04)	**1.05 (1.01–1.10)***
7 h	**1.07 (1.02–1.12)***	1.028 (0.99–1.08)	**1.06 (1.01–1.12)***	1.034 (0.99–1.09)	1.03 (0.98–1.08)	0.99 (0.94–1.04)	1.02 (0.98–1.08)
9 h	1.05 (0.99–1.10)	1.02 (0.97–1.70)	1.04 (0.98–1.09)	1.01 (0.96–1.06)	1.03 (0.98–1.07)	1.01 (0.96–1.06)	1.02 (0.97–1.08)
11.5 h	**1.10 (1.06–1.15)***	**1.07 (1.02–1.11)***	**1.10 (1.05–1.15)***	**1.08 (1.03–1.12)***	1.03 (0.98–1.07)	0.99 (0.95–1.04)	1.02 (0.98–1.07)
14.5 h	1.06 (0.10–1.12)	**1.06 (1.00–1.12)***	**1.08 (1.02–1.15)***	1.06 (1.00–1.12)	1.00 (0.96–1.06)	1.0 (0.96–1.06)	1.03 (0.97–1.09)
17 h	**1.07 (1.01–1.14)***	1.01 (0.95–1.07)	1.04 (0.98–1.11)	1.04 (0.98–1.10)	1.03 (0.97–1.10)	0.97 (0.91–1.03)	1.01 (0.95–1.06)
20 h	**1.08 (1.03–1.14)***	1.03 (0.98–1.08)	**1.07 (1.02–1.12)***	**1.05 (1.0–1.10)***	1.03 (0.98–1.08)	0.98 (0.93–1.03)	1.02 (0.97–1.07)
33 h	**1.05 (1.00–1.10)***	1.03 (0.98–1.08)	1.05 (0.99–1.09)	1.034 (0.99–1.09)	1.02 (0.97–1.07)	0.99 (0.95–1.04)	1.01 (0.97–1.06)

Data were analysed at each time point separately using a mixed-effect ANOVA model including fixed effects for treatment and period, a random effect for subjects, and baseline value (i.e. value observed at D0) as a covariate (posthoc analysis); and * bold values are signifiant différences $P < 0.05$.

CI, confidence interval; PVT, psychomotor vigilance test.

Treatments are: PBO, placebo modafinil + placebo flecainide; MOD100, modafinil 100 mg + placebo flecainide; THN102 100/1, modafinil 100 mg + 1 mg flecainide; THN102 100/3, modafinil 100 mg + 3 mg flecainide; and THN102 100/9, modafinil 100 mg + 9 mg flecainide. The data for treatments represent 12 subjects, except for THN102 100/9 that represent 11 subjects.

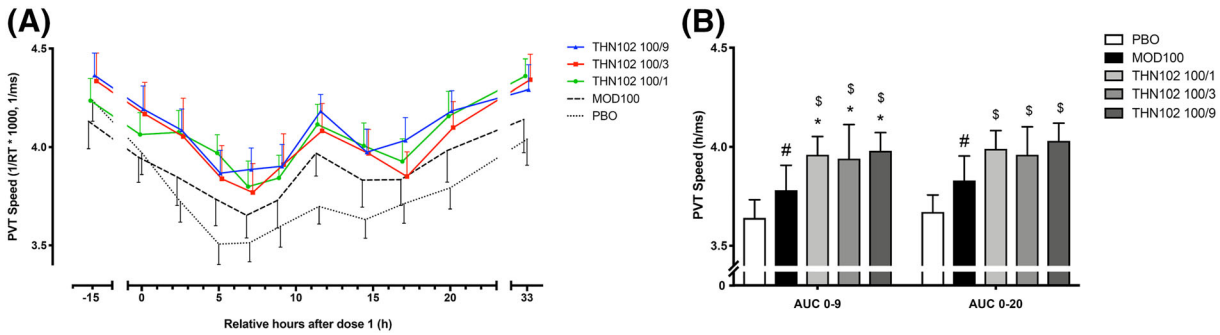

FIGURE 2 Reaction time (RT, 1/RT * 1000) in the PVT test at each time-point for the 3 doses THN102 (100/1, 100/3, and 100/9), modafinil alone (MOD100), and placebo (PBO) (A) and area under the curve between 0–9 hours and 0–20 hours post-treatment (B). Values are mean ± standard error. 100/1, 100/3, 100/9: combination of 100 mg modafinil with 1, 3 and 9 mg flecainide, respectively. PVT, psychomotor vigilance test. * THN102 100/1 *vs* MOD100 ($P < .05$), $ THN102 100/1 *vs* PBO ($P < .05$) and # MOD100 *vs* PBO ($P < .05$). Graphs in (A) are displaced slightly so that they are not graphically overlapping

and 100/9 only ($P < .05$). The variation for pick-up time in the ToL test with THN102 100/9 was significantly different *vs* MOD100 and PBO.

There was a significant period effect on the number of correct responses in the 2-Back task at baseline (on D0) and TSD (on D1; $P < .05$ for all).

Safety review

No serious adverse effects were reported. All AEs were rated mild to moderate, except 1 severe (somnolence). The most common drug-related AEs were fatigue, somnolence, headache, nausea, abdominal pain. No clinically relevant safety laboratory changes from baseline were observed in relation to treatments. No modification of ECG intervals (QTc, PR and QRS) was reported. Safety of THN102 (all doses) was similar to MOD100 alone, notably on somnolence (66 *vs* 75%), fatigue (70 *vs* 83%), headache (23 *vs* 50%), nausea (14 *vs* 33%; Table 4).

4 | DISCUSSION

In this study in healthy sleep-deprived subjects, we found that THN102, a combination of modafinil and low-dose flecainide, appears

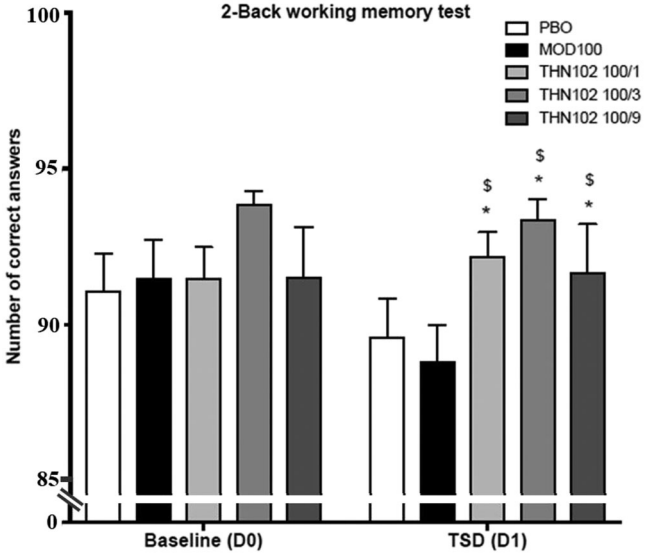

FIGURE 3 Number of correct responses in the 2-Back working memory test for the 3 doses THN102 (100/1, 100/3 and 100/9), modafinil alone (MOD100), and placebo (PBO)
* THN102 100/1, 100/3 and 100/9 *vs* MOD100 ($P < .05$), $ THN102 100/1, 100/3 and 100/9 *vs* PBO ($P < .05$), ns nonsignificant difference with placebo

superior to modafinil on attentional performance, working memory and executive functions. Adding flecainide low-dose boosts the wake-promoting effects of modafinil on cognitive functions altered during sleep deprivation, thus showing that the modulation of astroglial connexins by flecainide can increase the activity of modafinil. This is a first translation of this novel approach from animal studies to humans.

A cross-over design was selected to reduce sample size, as presented elsewhere in similar experimental sleep-deprivation conditions and evaluation of new drugs.[44] An incomplete 3-period cross-over design was selected to reduce the impact of a complete 5-period cross-over trial; a 4-week washout period was considered as sufficient to exclude pharmacological or pharmacokinetic carryover effect. Modafinil 100 mg was selected for its partial ability to restore attention during sleep deprivation,[11] and the treatment scheme (01:00, 09:00 and 19:00) as previously described.[45] Flecainide dose was determined using: (i) Food and Drug Administration allometric guidelines[46]; (ii) nonclinical pharmacokinetic studies; and (iii) pharmacokinetic determination from a previous unpublished clinical trial designed to assess THN102 safety. The flecainide doses of 1, 3 and 9 mg doses were then chosen. As flecainide had no effect when used alone in nonclinical models,[24] no flecainide-only treatment was included in this clinical study. In a previous clinical pharmacokinetic study and in this present study, pharmacokinetic data did not indicate that there was any pharmacokinetic interaction between modafinil and flecainide that could explain the increased activity on vigilance and cognition simply by an increase of modafinil exposure (data not shown).

For the treatment of tachyarrhythmia, flecainide is given at daily doses between 100 and 300 mg. Blood sampling in the present study demonstrated a mean maximal concentration of flecainide below 20 ng/mL for all groups (data not shown). This exposure level is well below exposure levels where cardiovascular AEs are expected for flecainide, as flecainide does not trigger any significant cardiovascular effects below 100 ng/mL or even 200 ng/mL in plasma.[47-50] The overall safety of THN102 was similar to modafinil; self-reported adverse events were equivalent between both treatments.

The PVT is a widely used measure of behavioural alertness[31] and PVT speed is considered as the most sensible parameter to total sleep deprivation.[32] In subjects receiving placebo, PVT speed rapidly decreased during sleep deprivation (40 hours of continuous wakefulness) to an early morning nadir at 06:00, consistent with published literature.[11,40] During sleep deprivation (54.5 hours), a single administration of 100 mg modafinil (MOD100) blunted the decrease of reaction time but did not completely restore the PVT performance.[11] In our study, we showed that, in subjects receiving MOD100, the PVT speed was significantly higher than placebo only at 5 and 11.5 hours after the start of the treatment. In comparison, the superiority of THN102 *vs* placebo is observed at 6 time points (2.5, 5, 7, 11.5, 14.5 and 20 hours after the first dose) during the protocol of sleep deprivation.

We demonstrated that a combination of a low dose of flecainide (1 mg) with modafinil improved the procognitive effect of modafinil alone in healthy subjects during sleep deprivation. In particular, we observed that THN102 100/1 resulted in a statistically higher PVT speed *vs* MOD at 5 hours postdose 1, and during the 9 first hours of treatment when expressed as AUC. Our results are partly consistent with a previous study in rodents showing that flecainide enhanced the wake-promoting and procognitive effects of modafinil.[24] In this study, the astroglial network mediated by connexins was suggested to have an impact on neuronal pathways controlling sleep–wake and cognitive capacities.

In our study, we demonstrated that all 3 THN102 doses significantly improved working memory (2-Back, number of correct responses) and part of executive process (Go errors in Go/noGo) *vs* modafinil during sleep deprivation. The perseverative errors in WCST were improved for THN102 100/1 and 100/9 only. In contrast, MOD100 alone had no effect on working memory and executive processes compared to placebo and could not prevent the decrease of performance. Previous studies showed a beneficial effect of modafinil on working memory in healthy young sleep deprived (2 nights) subjects but with 200 and 400 mg doses of modafinil, with a trend towards a dose-dependent effect.[11] In healthy young non–sleep-deprived subjects modafinil (100 and 200 mg) significantly enhances performance on planning, accuracy and inhibition tasks, mostly without dose-dependent effect.[51] The executive processes assessed using the WCST task were improved during sleep deprivation with 400 mg of modafinil[11-13,52] while no effect was observed in non–sleep-deprived subjects with 200 mg and 400 mg.[52] Go errors are typically considered as an indicator of lack of attention, while noGo errors responses are considered as indicators of impulsivity.[35] WCST is the classical test of executive function, i.e. test of set-shifting.[37] Apart from THN102 100/9, no effect of THN102 was observed on ToL score, potentially linked to the fact that inhibitory capacity and

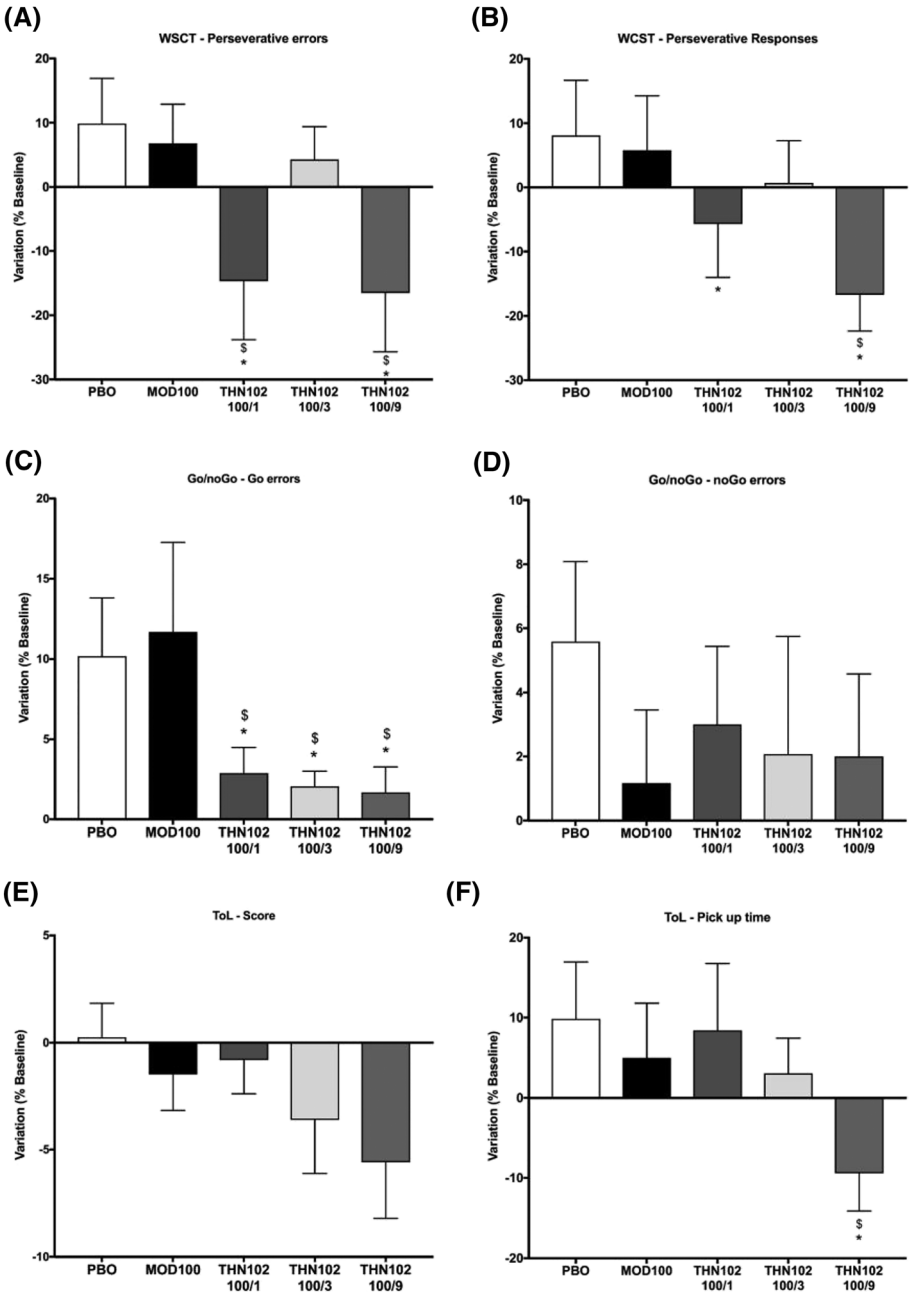

FIGURE 4 Variation (% baseline) in perseverative errors (A) and responses (B; flexibility) in the WCST test, in Go (C; inhibition) and noGo (D) errors in the Go/noGo test, and score (E) and pickup time (F; planning) in the ToL test for the 3 doses THN102 (100/1, 100/3, and 100/9), modafinil alone (MOD100), and placebo (PBO). WCST, Wisconsin card sorting test; ToL, Tower of London. * THN102 100/1, 100/3 and 100/9 *vs* MOD100 ($P < .05$), $ THN102 100/1, 100/3 and 100/9 *vs* PBO ($P < .05$)

working memory appear to contribute very little to the prediction of ToL scores.[53] Finally, in our study we did not observe significant difference between the 3 doses of THN102 (1, 3 and 9 mg of flecainide) on the primary objective psychomotor vigilance nor on working memory and executive inhibition processes, which suggests a flat dose–response in the tested conditions. Altogether, those data indicate the better efficacy of THN102 *vs* modafinil on vigilance and cognition impairments induced by sleep deprivation.

The study has several limitations that may be linked to the relative short wake-time (18 h) before the first administration of the tested drugs compared to reported studies with wake times around 44–65 hours.[11-13,54] This may explain the relatively good preservation of functions in this sample of healthy young male subjects decreasing the probability to detect contrasts between modafinil and THN102. Only male participants were included in the study for feasibility reasons (military environment), although sex-specific differences would not necessarily have been expected. This study has been performed on a limited number of subjects (for ethical, logistical and safety considerations linked to a first administration in sleep deprived healthy subjects), which could explain the lack of statistical significance

TABLE 4 Treatment-related key adverse events (AEs)

	PBO *n* = 12	MOD100 *n* = 12	THN102 100/1 *n* = 12	THN102 all doses *n* = 35
Somnolence	11 (92%)	9 (75%)	8 (67%)	23 (66%)
Fatigue	10 (83%)	10 (83%)	8 (67%)	24 (70%)
Abdominal pain	3 (25%)	2 (17%)	3 (25%)	6 (17%)
Headache	2 (17%)	6 (50%)	4 (33%)	8 (23%)
Nausea	2 (17%)	4 (33%)	4 (33%)	5 (14%)
Dry mouth	1 (8%)	0 (0%)	1 (8%)	3 (8%)

Values are *n* (%) with treatment related AEs. AEs are combination of spontaneous reporting and of VAS if >20/100 mm. Last column aggregates the total symptoms observed after all doses of THN102 (100/1 mg, 100/3 mg and 100/9 mg).

between THN102 and MOD100 at several time points, whereas AUC values are significantly different. As usual in pharmacodynamic studies, multiple outcomes were tested without correction for multiplicity, but the overall pattern of results indicates consistent advantages of the combination *vs* modafinil alone.

The tasks used in this study represent a small sample of the range of cognitive processes. In particular, future research should examine the effects on other dimensions of executive functions, such as the ability to update or rehearse,[53] in order to further explore the cognitive activity of THN102, as well as the impact after chronic treatment. A Phase II trial in narcoleptic patients is ongoing (NCT02821715), with evaluation of excessive daytime sleepiness as primary objective.

In conclusion, THN102 seems clinically superior to modafinil on vigilance, working memory and executive functions in healthy sleep deprived subjects. This result should be confirmed in a larger healthy population and patients. The enhancement of the alertness and cognitive outcomes of modafinil demonstrated here with flecainide would open new perspectives in the treatment of symptoms associated with hypersomnolence disorders (narcolepsy, Parkinson's disease, idiopathic hypersomnia etc.) such as the deficit of attention, executive processes and memory, or to alleviate the deleterious effects of wake–sleep rhythm disruptions during prolonged military operations on vigilance and cognitive performance.[6-9]

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COMPETING INTERESTS

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CONTRIBUTORS

F.S., F.M., F.B.-F., M.Che. and M.Cha conceived and planned the experiments. F.S., M.E., M.Che., D.G-M., A.R., C.Bour., M.G., H.D., H.L., C.D., P.V-B., C.Boug., P.A., and M.Che carried out the experiments. F.S., F.M., F.B.-F., D.G-M., M.Che., W.R., Y.D., D.L. and M. Cha contributed to the interpretation of the results. F.S. took the lead in writing the manuscript. All authors provided critical feedback and helped shape the research, analysis and manuscript.

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REFERENCES

1. Kornum BR, Knudsen S, Ollila HM, et al. Narcolepsy. *Nat Rev Dis Primers*. 2017;3(1):16100.
2. Barateau L, Lopez R, Dauvilliers Y. Treatment options for narcolepsy. *CNS Drugs*. 2016;30(5):369-379.
3. Avellar AB, Carvalho LB, Prado GF, Prado LB. Pharmacotherapy for residual excessive sleepiness and cognition in CPAP-treated patients with obstructive sleep apnea syndrome: a systematic review and meta-analysis. *Sleep Med Rev*. 2016;30:97-107.
4. Cheng YH, Roach GD, Petrilli R. Current and future directions in clinical fatigue management: an update for emergency medicine practitioners. *Emerg Med Australas*. 2014;26(6):640-644.
5. Högl B, Saletu M, Brandauer E, et al. Modafinil for the treatment of daytime sleepiness in Parkinson\' s disease: a double-blind, randomized, crossover, placebo-controlled Polygraphic trial. *Sleep*. 2002;25(8):62-66.
6. Naumann A, Bellebaum C, Daum I. Cognitive deficits in narcolepsy. *J Sleep Res*. 2006;15(3):329-338.
7. Delazer M, Högl B, Zamarian L, et al. Executive functions, information sampling, and decision making in narcolepsy with cataplexy. *Neuropsychology*. 2011;25(4):477-487.
8. Rogers AE, Rosenberg RS. Tests of memory in narcoleptics. *Sleep*. 1990;13(1):42-52.
9. Estrada A, Kelley AM, Webb CM, Athy JR, Crowley JS. Modafinil as a replacement for dextroamphetamine for sustaining alertness in military helicopter pilots. *Aviat Space Environ Med*. 2012;83(6):556-567.
10. Ray K, Chatterjee A, Panjwani U, et al. Modafinil improves event related potentials P300 and contingent negative variation after 24h sleep deprivation. *Life Sci*. 2012;91(3):94-99.
11. Wesensten N, Belenky G, Kautz MA, Thorne DR, Reichardt RM, Balkin TJ. Maintaining alertness and performance during sleep deprivation: modafinil *vs* caffeine. *Psychopharmacology (Berl)*. 2002;159(3):238-247.
12. Wesensten NJ, Killgore WD, Balkin TJ. Performance and alertness effects of caffeine, dextroamphetamine, and modafinil during sleep deprivation. *J Sleep Res*. 2005;14(3):255-266.
13. Killgore W, Kahn-Greene ET, Grugle NL, Killgore DB, Balkin TJ. Sustaining executive functions during sleep deprivation: a comparison of

caffeine, dextroamphetamine, and modafinil. *Sleep*. 2009;32(2):205-216.

14. Fry, US Modafinil in Narcolepsy Multicenter Study Group. Randomized trial of modafinil for the treatment of pathological somnolence in narcolepsy. *Ann Neurol*. 1998;43(1):88-97. https://doi.org/10.1002/ana.410430115
15. Dauvilliers Y, Paquereau J, Bastuji H, Drouot X, Weil JS, Viot-Blanc V. Psychological health in central hypersomnias: the French harmony study. *J Neurol Neurosurg Psychiatry*. 2009;80(6):636-641. https://doi.org/10.1136/jnnp.2008.161588
16. Bayard S, Langenier MC, De Cock VC, Scholz S, Dauvilliers Y. Executive control of attention in narcolepsy. *PLoS ONE*. 2012;7(4):e33525.
17. Minzenberg MJ, Carter CS. Modafinil: a review of neurochemical actions and effects on cognition. *Neuropsychopharmacology*. 2008;33(7):1477-1502.
18. Liu X, Petit J-M, Ezan P, Gyger J, Magistretti P, Giaume C. The psychostimulant modafinil enhances gap junctional communication in cortical astrocytes. *Neuropharmacology*. 2013;75:533-538.
19. Halassa MM, Florian C, Fellin T, et al. Astrocytic modulation of sleep homeostasis and cognitive consequences of sleep loss. *Neuron*. 2009;61(2):213-219. https://doi.org/10.1016/j.neuron.2008.11.024
20. Franco-Perez J, Ballesteros-Zebadua P, Fernandez-Figueroa EA, Ruiz-Olmedo I, Reyes-Grajeda P, Paz C. Sleep deprivation and sleep recovery modifies connexin36 and connexin43 protein levels in rat brain. *Neuroreport*. 2012;23(2):103-107. https://doi.org/10.1097/WNR.0b013e32834e8fcb
21. Franco-Perez J, Paz C. Quinine, a selective gap junction blocker, decreases REM sleep in rats. *Pharmacol Biochem Behav*. 2009;94(2):250-254. https://doi.org/10.1016/j.pbb.2009.09.003
22. Charvériat M, Naus CC, Leybaert L, Sáez JC, Giaume C. Connexin-dependent Neuroglial networking as a new therapeutic target. *Front Cell Neurosci*. 2017;11:174. https://doi.org/10.3389/fncel.2017.00174
23. Picoli C, Nouvel V, Aubry F, et al. Human connexin channel specificity of classical and new gap junction inhibitors. *J Biomol Screen*. 2012;17(10):1339-1347.
24. Duchêne A, Perier M, Zhao Y, et al. Impact of Astroglial Connexins on Modafinil pharmacological properties. *Sleep*. 2016;39(6):1283-1292.
25. Vodovar D, Duchêne A, Wimberley C, et al. Cortico-amygdala-striatal activation by Modafinil/Flecainide combination. *Int J Neuropsychopharmacol*. 2018;21(7):687-696. https://doi.org/10.1093/ijnp/pyy027
26. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. *Acta Psychiatr Scand*. 1983;67(6):361-370.
27. Johns MW. A new method for measuring daytime sleepiness: the Epworth sleepiness scale. *Sleep*. 1991;14(6):540-545.
28. Horne JA, Ostberg O. A self-assessment questionnaire to determine morningness-eveningness in human circadian rhythms. *Int J Chronobiol*. 1976;4(2):97-110.
29. Patterson H, Lucas H. Change-over designs. *North Carolina Agr Exp Sta Tech Bull*. 1962;147:52.
30. Ratkowsky D, Alldredge R, Evans MA. *Cross-over Experiments: Design, Analysis and Application* (Vol. 135). New York, USA: CRC Press; 1992.
31. Khitrov MY, Laxminarayan S, Thorsley D, et al. PC-PVT: a platform for psychomotor vigilance task testing, analysis, and prediction. *Behav Res Methods*. 2014;46(1):140-147.
32. Basner M, Dinges DF. Maximizing sensitivity of the psychomotor vigilance test (PVT) to sleep loss. *Sleep*. 2011;34(5):581-591.
33. Mueller ST, Piper BJ. The psychology experiment building language (PEBL) and PEBL test battery. *J Neurosci Methods*. 2014;222:250-259.
34. Braver TS, Cohen JD, Nystrom LE, Jonides J, Smith EE, Noll DC. A parametric study of prefrontal cortex involvement in human working memory. *Neuroimage*. 1997;5(1):49-62.
35. Bezdjian S, Baker LA, Lozano DI, Raine A. Assessing inattention and impulsivity in children during the go/NoGo task. *Br J Dev Psychol*. 2009;27(2):365-383.
36. Shallice T. Specific impairments of planning. *Philos Trans R Soc Lond B Biol Sci*. 1982;298(1089):199-209.
37. Monchi O, Petrides M, Petre V, Worsley K, Dagher A. Wisconsin card sorting revisited: distinct neural circuits participating in different stages of the task identified by event-related functional magnetic resonance imaging. *J Neurosci*. 2001;21(19):7733-7741.
38. Roth T, Schwartz JR, Hirshkowitz M, Erman MK, Dayno JM, Arora S. Evaluation of the safety of modafinil for treatment of excessive sleepiness. *J Clin Sleep Med JCSM Off Publ Am Acad Sleep Med*. 2007;3(6):595.
39. Arnal PJ, Sauvet F, Leger D, et al. Benefits of sleep extension on sustained attention and sleep pressure before and during Total sleep deprivation and recovery. *Sleep*. 2014;38(12):1935-1943.
40. Chennaoui M, Sauvet F, Drogou C, et al. Effect of one night of sleep loss on changes in tumor necrosis factor alpha (TNF-α) levels in healthy men. *Cytokine*. 2011;56(2):318-324.
41. Brown H, Prescott R. *Applied Mixed Models in Medicine*. Hoboken, New Jersey, USA: John Wiley & Sons; 2014.
42. Littell RC, Milliken GA, Stroup WW, Wolfinger RD, Schabenberger O. *SAS for Mixed Models*. 2 NC: SAS institute Cary; 2006.
43. Olofsen E, Dinges DF, Van Dongen H. Nonlinear mixed-effects modeling: individualization and prediction. *Aviat Space Environ Med*. 2004;75(3):A134-A140.
44. Iannone R, Palcza J, Renger JJ, et al. Acute alertness-promoting effects of a novel histamine subtype-3 receptor inverse agonist in healthy sleep-deprived male volunteers. *Clin Pharmacol Ther*. 2010;88(6):831-839. https://doi.org/10.1038/clpt.2010.205
45. Lagarde D, Batejat D, Van Beers P, Sarafian D, Pradella S. Interest of modafinil, a new psychostimulant, during a sixty-hour sleep deprivation experiment. *Fundam Clin Pharmacol*. 1995;9(3):271-279.
46. Guidance for Industry Estimating the Maximum Safe, Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers. https://www.fda.gov/downloads/drugs/guidances/ucm078932.pdf.
47. Funck-Brentano C, Becquemont L, Kroemer HK, et al. Variable disposition kinetics and electrocardiographic effects of flecainide during repeated dosing in humans: contribution of genetic factors, dose-dependent clearance, and interaction with amiodarone. *Clin Pharmacol Ther*. 1994;55(3):256-269.
48. Beckmann J, Hertrampf R, Gundert-Remy U, Mikus G, Gross AS, Eichelbaum M. Is there a genetic factor in flecainide toxicity? *BMJ*. 1988;297(6659):1316.
49. Salerno DM, Granrud G, Sharkey P, et al. Pharmacodynamics and side effects of flecainide acetate. *Clin Pharmacol Ther*. 1986;40(1):101-107.
50. Pritchett EL, DaTorre SD, Platt ML, McCarville SE, Hougham AJ. Flecainide acetate treatment of paroxysmal supraventricular tachycardia and paroxysmal atrial fibrillation: dose-response studies. The Flecainide supraventricular tachycardia study group. *J Am Coll Cardiol*. 1991;17(2):297-303.
51. Turner DC, Robbins TW, Clark L, Aron AR, Dowson J, Sahakian BJ. Cognitive enhancing effects of modafinil in healthy volunteers. *Psychopharmacology (Berl)*. 2003;165(3):260-269.

52. Cope ZA, Minassian A, Kreitner D, et al. Modafinil improves attentional performance in healthy, non-sleep deprived humans at doses not inducing hyperarousal across species. *Neuropharmacology*. 2017;125:254-262.

53. Smith EE, Jonides J. Storage and executive processes in the frontal lobes. *Science*. 1999;283(5408):1657-1661.

54. Wesensten NJ, Belenky G, Thorne DR, Kautz MA, Balkin TJ. Modafinil *vs* caffeine: effects on fatigue during sleep deprivation. *Aviat Space Environ Med*. 2004;75(6):520-525.

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